**Molecular characterization of Vibrio harveyi virulence associated serine protease and outer membrane protein genes for vaccine development**

ABSTRACT

Vibrio harveyi is the most pathogenic species associated with the infection in a wide range of marine species in aquaculture industries. A few virulence-associated genes have been discovered in V. harveyi. This study reports the cloning, sequence analysis and phylogenetic study of serine protease (VHS) and outer membrane protein (OMP) from a pathogenic V. harveyi, which isolated from a local outbreak of diseased tiger grouper. Molecular identification revealed that VHS and OMP consist of 1368 and 816 base pairs and encoded for mature peptides of 429 and 251 amino acids, respectively. The amino acids sequence identities of VHS was 100% similarity with protease of V. harveyi and OMP was 99% of membrane protein of V. harveyi, as compared to published sequence. Phylogenetic analysis and conserved domain search proposed that VHS is a serine endoprotease DegQ and OMP is an OmpK type. Signal peptide, transmembrane β-barrel and subcellular localization have supported the findings whereby demonstrated VHS belonged to periplasmic serine protease DegQ, composed one β-barrel and two α-barrels region. OMP displayed six β-barrels and twelve α-barrels suggesting it is belonged to outer membrane integral membrane protein, specifically act as a porin type outer membrane protein. Prediction of antigenic sites revealed that VHS composed 62 sites and OMP have 36 antigenic sites, assuming that they can provoke immune response of the infected hosts. In conclusion, it is strongly suggests that both genes can be potentially used for developing an effective live-attenuated vaccine candidate against vibriosis and further be applied in aquaculture industries.

**Keyword:** Molecular characterization; Vibrio harveyi; Virulence-associated; Serine protease; Outer membrane protein; Vaccine development